
The role of nutrients in cyanobacterial blooms in a shallow reservoir

Stefanie Mueller (Dipl.-Umweltwiss.)

A thesis submitted in fulfilment
of the requirements for the degree of
Doctor of Philosophy

School of the Environment
University of Technology, Sydney

April 1st, 2014

Certificate of original authorship

I certify that the work in this thesis has not previously been submitted for a degree nor has it been submitted as part of requirements for a degree except as fully acknowledged within the text.

I also certify that the thesis has been written by me. Any help that I have received in my research work and the preparation of the thesis itself has been acknowledged. In addition, I certify that all information sources and literature used are indicated in the thesis.

Signature

Date

Acknowledgments

I have been looking forward to writing this section for several months now as I felt that it would mark the completion of my thesis and I should save it up for the very end when every graph would have been plotted, every chapter written and every section typeset. Having now reached this point and looking back, I feel grateful to have had the opportunity to do a PhD thesis in the first place.

I would not have come this far had I not had help, encouragement and support from many others, first and foremost my supervisors: Simon Mitrovic and Darren Baldwin.

Simon, your input from inception to completion of my thesis has been invaluable. In particular, our fruitful discussions over many cups of good coffee and your ideas and feedback on manuscripts for reports, conference presentations, publications and of course this thesis have been tremendously helpful. Thank you for your encouragement and ongoing support over the last five years.

Darren, I owe you many thanks for your invaluable ideas and expert comments. I appreciate it very much that you have been very approachable despite the geographical distance between Albury and Sydney, not to mention generous with your time and your amazing cooking skills.

I would also like to give special thanks to Richard Lim who was instrumental in providing me with the opportunity to do this PhD.

I would like to thank Hunter Water who have made this project possible from the financial viewpoint. Special thanks go to Bruce Cole for organising the funding for this thesis and providing access to Hunter Water monitoring data. Ian Graham, Stuart Bailey, Jim Carter, Greg Mason and Paul have played a vital part in my fieldwork, they deserve a big thank you for patiently chauffeuring me all over Grahamstown Dam and showing an active interest in my experiments. I sincerely hope this thesis will provide some useful information for the management of the lake.

I feel like I have learned a lot during the work for this thesis thanks to many great teachers. At UTS, Gemma Armstrong and Sue Fenech have spared many hours teaching me the intricate workings of the flow injection analyser and the TCN analyser and have always been extremely helpful in all small and big issues

arising during one's everyday lab work. I am very grateful to Anne Colville who did a huge chunk of the work for the first *Anabaena circinalis* experiment, let alone the culturing of the cyanobacterium for many years now. Moreover, Anne has always been generous with providing equipment and useful advice. Thank you to Peter Jones who has been incredibly helpful in finding all sorts of laboratory and field equipment in the UTS stores and advising on its proper use.

I owe thanks to Jon Holiday from the Office of Water for teaching me one of the key skills in this endeavour: to correctly identify the freshwater phytoplankton of Grahamstown Dam to genus level. I am grateful to Jesko Strala for introducing me to L^AT_EX many years ago. It has actually made the typesetting of this thesis fun. Similarly, I need to thank Craig Syms for his introductory course in R which not only inspired me to start using the open source software but also to learn a lot more about statistical methods. Regarding the latter, Fraser Thorpe's expertise has been essential, too.

Further generous help for fieldwork came from James Hitchcock, Martin Pfeil and of course Simon Mitrovic. James, the Hitchcock trap was a great success!

Aside from field work, my dear husband Martin deserves acknowledgement for his immovable trust in my ability to complete this thesis even when I was critical with everything including myself and for enduring my absent mindedness while writing.

I never thought that a shared office could be a productive environment but I am glad I was wrong! I have very much enjoyed the company of my office mates Ann-Marie Rohlfs, Louisa Norman, Bec Herron, James Hitchcock, Lloyd Werry and Carolina Lopez. I will miss our exchange about housemates, statistics, cats, cake, black milk, experiments and many other things that kept us going.

Ganz herzlichen Dank, to my parents and grandma in Germany for all your supportive emails, phone calls and letters and to my German speaking friends all over the globe, for putting up with my absence on skype or email. My MUWi girls Anja, Jessi, Cordu and Friedi, you have been an inspiration – übrigens ist mir schon wieder kalt! A big thank you to my friends in Australia, especially Su Li and Brent, for cheering me on and for much needed distractions on many good climbing trips.

Contents

List of Figures	ix
List of Tables	xv
Abstract	xix
1 Introduction	1
1.1 Scope and need for this study	1
1.2 General features of cyanobacteria	3
1.2.1 Description of cyanobacteria	3
1.2.2 Bloom formation	5
1.2.3 Impact of cyanobacterial blooms on human health	5
1.3 Eutrophication, cyanobacteria and water management	8
1.4 Nutrient influence on cyanobacterial growth	11
1.4.1 Nutrient limitation	11
1.4.2 Investigating nutrient limitation	13
1.4.3 Competition for nutrients and cyanobacterial dominance	15
1.4.4 Nutrient inflow events	19
1.4.5 Nutrient release from sediments	20
1.5 Management of cyanobacteria	25
1.5.1 Nutrient management	25
1.5.2 Inflow management	29
1.5.3 Stratification management	30
1.6 Aims and overview of this study	31
2 Nutrient limitation in Grahamstown Dam	35

Contents

2.1	Introduction	35
2.2	Methods	37
2.2.1	Study site	37
2.2.2	Seasonal microcosm enrichment assays	40
2.2.3	Mesocosm enrichment assays	41
2.2.4	Data analysis	42
2.3	Results	43
2.3.1	Seasonal microcosm enrichment assays	43
2.3.2	Mesocosm enrichment assays	48
2.4	Discussion	54
3	Effects of light and trace metals on cyanobacterial growth in Grahams-	
	town Dam	62
3.1	Introduction	62
3.2	Methods	64
3.2.1	Data analysis	66
3.3	Results	67
3.3.1	Light level assay	67
3.3.2	Micronutrient assay	72
3.4	Discussion	76
4	Assessing the importance of the nutrients N and P for the growth of	
	<i>Anabaena circinalis</i>	80
4.1	Introduction	80
4.2	Methods	82
4.2.1	<i>A. circinalis</i> cultures	82
4.2.2	Batch experiments	84
4.2.3	Data analysis	87
4.3	Results	87
4.4	Discussion	93
5	Nutrient release from the sediments in Grahamstown Dam	98
5.1	Introduction	98
5.2	Methods	100

5.2.1	Sediment incubation experiments	100
5.2.2	Thermal stratification	101
5.2.3	Data analysis	102
5.3	Results	102
5.3.1	Thermal stratification	102
5.3.2	Environmental conditions in the incubation chambers	102
5.3.3	Nutrient release from the sediments	104
5.4	Discussion	110
6	Effects of organic carbon on sediment nutrient release and sediment characteristics	116
6.1	Introduction	116
6.2	Methods	118
6.2.1	Carbon limitation experiment	118
6.2.2	Sediment composition	119
6.3	Results	120
6.3.1	Environmental conditions in the incubation chambers	120
6.3.2	Nutrient release from the sediments	121
6.3.3	Sediment composition	123
6.4	Discussion	125
7	General discussion and conclusion	132
7.1	Summary and discussion	132
7.1.1	Recommendations for future research	139
7.1.2	Implications for management of algal and cyanobacterial growth in Grahamstown Dam	140
7.2	Conclusion	141
	Bibliography	143
A	Nutrient limitation in Grahamstown Dam	177
B	Effects of light and trace metals on cyanobacterial growth in Grahamstown Dam	180

C	Assessing the importance of the nutrients N and P for the growth of <i>Anabaena circinalis</i>	182
D	Nutrient release from the sediments in Grahamstown Dam	185

List of Figures

2.1	Location of Grahamstown Dam on the Australian continent and location of the three experimental sites within the lake. Solid circles mark approximate positions.	38
2.2	Biweekly monitoring of nitrate (NO_3) and srP at Seaham Weir on the Williams River and at the monitoring site close to Balickera Canal in Grahamstown Dam. Data presented were collected between 1/04/2000 and 1/04/2009 by Hunter Water.	39
2.3	Chlorophyll a concentration for all three sites during the seasonal microcosm nutrient enrichment assays conducted from April 2009 until August 2011. Treatments with N only addition (N), P only addition (P) and N and P added in combination (PN) and the control (C) display values on day 4. Initial concentrations (i) were measured on day 0 prior to nutrient additions. Error bars are one standard error from the mean, n=3.	44
2.4	Chlorophyll a concentration in $\mu\text{g/L}$ in the mesocosm and microcosm assays conducted in January and March 2013. Error bars are one standard error from the mean, n=3.	48
2.5	Biovolumes in mm^3/L of all phytoplankton genera responding to additions of P and N in combination in the mesocosm assays in January 2013. Error bars are one standard error from the mean, n=3.	50
2.6	Biovolumes in mm^3/L of all phytoplankton genera responding to additions of P and N in combination in the mesocosm assays in March 2013. Error bars are one standard error from the mean, n=3.	52

- 2.7 Biovolumes in mm^3/L of phytoplankton genera responding to additions of P or N and genera switching responses to different treatments. Biovolumes of *Anabaena* and *Aphanizomenon* have been square root transformed because of low values at the beginning of the experiment. Graphs not further labeled show responses of genera in the mesocosm assay in January 2013. Error bars are one standard error from the mean, $n=3$ 53
- 3.1 Mean biovolumes in the light level assay. Letters a and b indicate homogeneous subsets according to the result of Tukey's pairwise comparison ($p=0.029$). Initial values (samples from day 0) were not included in the Tukey's test. Error bars are one standard error from the mean, $n=3$ 67
- 3.2 NMDS plot of the most abundant phytoplankton genera in the light level assay. Species scores (phytoplankton genera) are weighted and expanded averages of site scores (treatments) which leads to species scores being close to the site scores they are relatively more associated with. Names of genera were abbreviated and stand for Anab = *Anabaena*, Aphaniz = *Aphanizomenon*, Aphanoc = *Aphanocapsa*, Chrooc = *Chroococcus*, Ankistro = *Ankistrodesmus*, Choda = *Chodatella*, Clos = *Closterium*, Cruci = *Crucigenia*, Dictyo = *Dictyosphaerium*, Elaka = *Elakatothrix*, Mouge = *Mougeotia*, Nephro = *Nephrocystium*, Ooc = *Oocystis*, Pedi = *Pediastrum*, Scene = *Scenedesmus*, Sphaero = *Sphaerocystis*, Staurast = *Staurastrum*, Tetra = *Tetradron*, Acanth = *Acanthoceras*, Aulaco = *Aulacoseira*, Cyclo = *Cyclotella*, Syne = *Synedra*, Uroso = *Urosolenia*, Eugl = *Euglena*, Trachelo = *Trachelomonas*, Chroo = *Chroomonas*, Crypto = *Cryptomonas* and Dinob = *Dinobryon*. . . . 70
- 3.3 Mean chlorophyll a concentration in the light level assay. Letters a, b and c indicate homogeneous subsets according to the result of Tukey's pairwise comparison ($p \leq 0.001$). Initial values (samples from day 0) were not included in the Tukey's test. Error bars are one standard error from the mean, $n=3$ 71

- 3.4 Mean biovolumes in the micronutrient assay. Letters a and b indicate homogeneous subsets according to the result of Tukey's pairwise comparison ($p=0.029$). Initial values (samples from day 0) were not included in the Tukey's test. Error bars are one standard error from the mean, $n=3$ (except for treatment "N trace metals added" where $n=2$). 72
- 3.5 NMDS plot of the most abundant phytoplankton genera in the micronutrient assay. Species scores (phytoplankton genera) are weighted and expanded averages of site scores (treatments) which leads to species scores being close to the site scores they are relatively more associated with. Names of genera were abbreviated and stand for Anab = *Anabaena*, Aph-aniz = *Aphanizomenon*, Aphanoc = *Aphanocapsa*, Chrooc = *Chroococcus*, Ankistro = *Ankistrodesmus*, Choda = *Chodatella*, Clos = *Closterium*, Cruci = *Crucigenia*, Dictyo = *Dictyosphaerium*, Elaka = *Elakatothrix*, Mouge = *Mougeotia*, Nephro = *Nephrocystium*, Ooc = *Oocystis*, Pedi = *Pediastrum*, Scene = *Scenedesmus*, Sphaero = *Sphaerocystis*, Staurast = *Staurastrum*, Tetra = *Tetraedron*, Acanth = *Acanthoceras*, Aulaco = *Aulacoseira*, Cyclo = *Cyclotella*, Syne = *Synedra*, Uroso = *Urosolenia*, Eugl = *Euglena*, Trachelo = *Trachelomonas*, Chroo = *Chroomonas*, Crypto = *Cryptomonas* and Dinob = *Dinobryon*. $n=3$, except for treatment "N trace metals added" where $n=2$ 74
- 3.6 Mean chlorophyll a concentration in the micronutrient assay. Letters a, b, c, d and e indicate homogeneous subsets according to the result of Tukey's pairwise comparison ($p \leq 0.012$). Initial values (samples from day 0) were not included in the Tukey's test. Error bars are one standard error from the mean, $n=3$ (except for treatment "N trace metals added" where $n=2$). 76
- 4.1 Growth of *A. circinalis* in the first batch experiment. Note that the y-axis has a logarithmic scale. Error bars are one standard error from the mean, $n=3$ 88
- 4.2 *A. circinalis* growth rate per day and yield (cells/ml) for all nutrient treatments with positive growth and the control (C) in the first batch experiment. Error bars are one standard error from the mean, $n=3$ 89

4.3	Ratio of heterocysts to vegetative cells of <i>A. circinalis</i> in the first batch experiment. Error bars are one standard error from the mean, n=3.	90
4.4	Growth of <i>A. circinalis</i> measured as optical density (OD) in the second batch experiment. Error bars are one standard error from the mean, n=3. .	91
4.5	Mean growth rates in the second batch experiment. Error bars are one standard error from the mean, n=3 (except for treatment 40 N1P where n=1).	92
4.6	Mean yield measured as maximum optical density (OD) in the second batch experiment. Error bars are one standard error from the mean, n=3. .	92
5.1	Diagram of a sediment incubation chamber (used for the anoxic treatments) consisting of an airtight plastic box fitted with a butyl rubber septum as a sampling port and a nitrile glove connected to the outside of the box for pressure equalisation during sampling.	101
5.2	Temperature profile of the water column at Site 2 in Grahamstown Dam and temperature difference between surface and 9 m during the period of the most pronounced stratification between September 2011 and February 2013. Shaded areas highlight temperature differences >0.15 °C (according to Sherman et al. (1998), persistent thermal stratification occurs when the difference between water surface and bottom water is >0 °) and lasting for more than 24 hours. Data was smoothed with a rolling mean with a window of 10 observations.	103
5.3	N and P species and Fe(II) concentration in both treatments during the sediment release experiment in winter 2010. Concentrations are averages of five replicates from the three sites. Note that the graph for Fe(II) has a logarithmic y-axis. Error bars are one standard error from the mean, n=15, except for srP where n=12.	105
5.4	N and P species and Fe(II) concentration in both treatments during the sediment release experiment in summer 2011. Concentrations are averages of five replicates from the three sites. Note that the graph for Fe(II) has a logarithmic y-axis. Error bars are one standard error from the mean, n=15.	106

5.5	NMDS ordination plots for all nutrient data in the sediment nutrient release experiments conducted in winter and summer. Species scores ($\text{NH}_4^+=\text{NH}_4$, $\text{NO}_x=\text{Nox}$, TN, srP, TP, Fe) are weighted and expanded averages of sample scores (species and sample scores have equal variances) which leads to species scores being close to the sample scores they are relatively more associated with. Convex hulls indicate sample scores belonging to the three sites and the anoxic (extended to the right) and oxic (on the left) treatments.	107
6.1	Nutrients released over the 28 day C limitation experiment. Error bars are one standard error from the mean, n=4.	122
A.1	Nutrient concentrations in $\mu\text{g/L}$ in the mesocosm assays. Error bars are one standard error from the mean, n=3.	179
B.1	Profile of the light intensity measured at the beginning of the light level assay. The line represents the result of a nonlinear regression (performed with the nls function of the statistics software R) of the measured values ($R^2=0.96$). Depth corresponding to 10, 25 and 90 % light intensity were calculated with the model fitted in the nonlinear regression.	181
C.1	Calibration curve of optical density (OD) and cell counts. OD was measured at 560 nm.	184
D.1	Temperature profile of the water column at Site 1 in Grahamstown Dam. Data was smoothed with a rolling mean with a window of 100 observations. Temperature loggers at 0.5, 1 and 7 m stopped recording on the 2/11/2012. The shaded area represents a period with pronounced thermal stratification. Data from that periods is shown in more detail in Figure D.4.	186

- D.2 Temperature profile of the water column at Site 2 in Grahamstown Dam. Data was smoothed with a rolling mean with a window of 100 observations. The temperature logger at 8.5 m was accidentally detached when downloading data on the 1/11/2012 and reattached on the 27/11/2012. Data recorded between those dates is not shown. The shaded area represents a period with pronounced thermal stratification. Data from that periods is shown in more detail in Figure 5.2. 187
- D.3 Temperature profile of the water column at Site 3 in Grahamstown Dam. Data was smoothed with a rolling mean with a window of 100 observations. The shaded area represents a period with pronounced thermal stratification. Data from that periods is shown in more detail in Figure D.5. 188
- D.4 Temperature profile of the water column and temperature differences between different water depths at Site 1 in Grahamstown Dam during the period of the most pronounced stratification between September 2011 and February 2013. Data was smoothed with a rolling mean with a window of 48 observations. Shaded areas highlight temperature differences larger than 0.15°C and lasting for more than 24 hours. Data was smoothed with a rolling mean with a window of 10 observations. 189
- D.5 Temperature profile of the water column and temperature differences between different water depths at Site 3 in Grahamstown Dam during the period of the most pronounced stratification between September 2011 and February 2013. Data was smoothed with a rolling mean with a window of 48 observations. Shaded areas highlight temperature differences larger than 0.15°C and lasting for more than 24 hours. Data was smoothed with a rolling mean with a window of 10 observations. 190

List of Tables

2.1	Result of repeated measures ANOVA of the chlorophyll a response to the four nutrient treatments (C, N, P and PN) at the three sites in seven microcosm assays (time).	45
2.2	Growth responses of the phytoplankton genera that responded to the nutrient enrichment treatments for each site and each assay. Treatment names given in the table indicate which treatments evoked enhanced growth and asterisks show whether the growth response was significantly higher compared with the control. Superscript a indicate that there was no significant difference between marked treatments. No entry for any genus means there was no distinct growth in any of the treatments compared with controls. P values were derived from fully factorial two way ANOVA of site and nutrient enrichment treatment for each assay followed by Tukey's Pairwise Comparison.	46
2.3	Results of the comparison of the chlorophyll a concentration of the mesocosm and microcosm assays on day 4. Factor size had the levels microcosm and mesocosm, factor treatment had the levels C, N, P and PN in the two factorial ANOVA.	49
3.1	Elemental concentrations of trace metal solutions in the micronutrient assays. Stock solutions are from the Swedish Standard (SIS) <i>Lemna</i> growth medium (OECD, 2006).	66
3.2	Results of two factorial ANOVA of cyanobacterial biovolumes from the light level assay. Factor light had the levels 90 and 25 % surface irradiance and factor macronutrients had the levels C, N, P and PN. Initial values (samples from day 0) were not included in the analysis.	68

3.3	Results of PERMANOVA of the light level assay. Factor micronutrients had the levels trace metals present and trace metals absent and factor macronutrients had the levels C, N, P and PN. Number of permutations was 999. Initial values (samples from day 0) were not included in the analysis.	69
3.4	Results of two factorial ANOVA of chlorophyll a data from the light level assay. Factor light had the levels 90 and 25 % surface irradiance and factor macronutrients had the levels C, N, P and PN. Initial values (samples from day 0) were not included in the analysis.	71
3.5	Results of two factorial ANOVA of cyanobacterial biovolumes from the micronutrient assay. Factor micronutrients had the levels trace metals present and trace metals absent and factor macronutrients had the levels C, N, P and PN, n=3 (except for treatment "N trace metals added" where n=2). Initial values (samples from day 0) were not included in the analysis.	73
3.6	Results of PERMANOVA of the micronutrient assay. Factor micronutrient had the levels trace metals present and trace metals absent and factor macronutrient had the levels C, N, P and PN. Number of permutations was 999, n=3 (except for treatment "N trace metals added" where n=2). Initial values (samples from day 0) were not included in the analysis. . .	75
3.7	Results of two factorial ANOVA of chlorophyll a data from the micronutrient assay. Factor micronutrients had the levels trace metals present and trace metals absent and factor macronutrients had the levels C, N, P and PN, n=3 (except for treatment "N trace metals added" where n=2). Initial values (samples from day 0) were not included in the analysis.	76
4.1	Experimental design of the first batch experiment. All treatments and the control (C) were prepared in quadruplicate.	84
4.2	Nutrient concentrations in treatments in the second batch experiment. Nutrient treatments were tested at three different light levels with irradiances of 12, 40 and 95 $\mu\text{molm}^{-2}\text{s}^{-1}$. All treatments were prepared in triplicate.	85

4.3	Results of ANOVA of growth rate and yield from the first batch experiment. Factor nutrients had 14 levels (N3P1, N3P2, N3P3, N2P1, N2P2, N2P3, N1P1, N1P2, N1P3, N1, N2, N3 and C).	88
4.4	Nutrient concentrations of test medium at the end of the first batch experiment (day 14), intended concentration for each treatment on day 0 and concentration in the surrogates after seeding on d 0.	90
4.5	Results of ANOVA of growth rate and maximum optical density (OD) in the second batch experiment. For the growth rate, factor light had the levels 95, 40 and $12 \mu\text{molm}^{-2}\text{s}^{-1}$ and factor nutrients had the levels N2P and N1P. For maximum OD, light had the same levels as for the growth rate but nutrients had the levels N2P, N1P, N1 and P.	92
4.6	Nutrient concentrations and standard errors (s.e.) in surrogate treatments in the second batch experiment on day 0 before and after seeding with washed cell suspension. All treatments were prepared in triplicate.	93
4.7	Nutrient concentrations and standard errors (s.e.) in the second batch experiment on day 12. All treatments were prepared in triplicate.	93
6.1	Proportion and standard error (s.e.) in mg analyte per gram sediment based on dry weight. Pore water nutrient concentrations are in mg/L. TC_{ox} , TN_{ox} and TP_{ox} were measured in sediments collected from oxic incubation chambers after the winter nutrient release experiment (chapter 5). TC, TN and TP were measured in sediments collected in April 2013. LOI was measured as part of the XRF analysis (LOI_{XRF}) and by dry combustion at 550°C in samples from October 2011 (LOI). Elemental proportions of Al, Fe, Ca and P were calculated from Al_2O_3 , Fe_2O_3 , CaO and P_2O_5 values.	124
A.1	Mean nutrient concentrations in $\mu\text{g/L} \pm$ standard errors ($n=3$) in all nutrient enrichment assays measured on day 0 and day 4. Initial concentration (on day 0) were measured in samples taken from the filtered bulk water the experimental bottles were filled with. Amended nutrients (treatment PN on day 0) were measured in surrogate experimental bottles.	178
C.1	Swedish Standard (SIS) <i>Lemma</i> growth medium (OECD, 2006).	183

C.2	Base mixture for preparation of <i>Lemna</i> base medium (LM-N&P) used for batch experiments.	184
-----	--	-----

Abstract

This thesis examines potential causes for algal and cyanobacterial blooms in Grahamstown Dam, a shallow mesotrophic drinking water reservoir in coastal NSW, Australia. The objective was to understand the role of nitrogen and phosphorus in algal and cyanobacterial growth and to elucidate other chemical and physical processes that may enhance cyanobacterial growth in the lake.

Algal and cyanobacterial nutrient limitation was examined on different spatial and temporal scales in in situ assays. Other aspects that have been found to promote cyanobacteria, i.e. high irradiance levels as may occur during thermal stratification and trace metal nutrient additions, were investigated in situ. The effects of different nutrient supply ratios and different light climates on growth rate and yield of the prominent potentially toxic cyanobacterium *Anabaena circinalis* were tested in laboratory experiments. Different aspects of nutrient release from the sediments were examined under conditions that may occur during persistent thermal stratification, i.e. bottom water anoxia. Further experiments elucidated the influence of organic substrate on microbially mediated nutrient release process in the sediments.

Phytoplankton biomass and most individual genera were colimited by nitrogen and phosphorus. Further, the growth response of potentially toxic cyanobacteria lagged behind the response of most other phytoplankton. Many algae responded with increased growth to the combination of high irradiance and nutrient enrichment. The response of potentially toxic cyanobacteria was inconclusive. Trace metal nutrient additions enhanced the growth of one potentially toxic cyanobacterium and most non toxic genera. Nitrogen concentration and not nutrient ratio or phosphorus concentration determined yield of *A. circinalis*. This effect was increased by higher irradiance levels. Growth rates were enhanced

by high irradiance and high N concentration. The sediments were a source of N under oxic and anoxic conditions. Small amounts of phosphorus were released during anoxia only when the availability of dissolved organic C was improved, indicating microbiological activity as the cause of phosphorus release. Moreover, iron and phosphorus release was not caused by the same processes in the sediments.

These findings imply that a pulse of nutrients is not likely to lead to cyanobacterial blooms in Grahamstown Dam but it cannot be excluded that a gradual increase in nutrient load would not. Persistent thermal stratification may increase the risk of cyanobacterial growth by providing increased levels of nitrogen and an improved light climate. Unexpected results, such as insensitivity of cyanobacteria to nutrient enrichment, phytoplankton colimitation and decoupling of iron and phosphorus cycling in the sediments suggest that further research on shallow coastal lakes would be useful.